

Building Active Atlas for Mouse Brainstem

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Abstract. We propose a semi-supervised system for generating a reference atlas from histology image series of the mouse brainstem. The atlas contains the mean and variance of the positions and shapes of nine anatomical landmarks. By learning landmark classifiers, the system registers new data to the atlas. Registered data further improve the generalizability of the classifiers.

Keywords: landmark detection, atlas generation, mouse brain, registration, automated annotation

1 Overview

As one of the most important tools to neuroscientists, a brain atlas provides a standard model for the anatomy of a certain species' brain. It guides stereotactic surgery and recording of neural activities. It helps one establish the identity of a region of interest in a new sample. It also allows one to map different samples to a common coordinate system. This forms a skeleton for compiling new data, including tracing studies of neuronal connections, electrophysiological recording studies of the action of neurons in different areas, and anatomical studies of different cell types. With these, one can quantify the spatial variance of a particular structure between individuals, or correlate gene expression patterns spatially to discover neural circuitry.

In this paper we focus on building an atlas for the mouse brainstem. Existing mouse brain atlases, such as the Paxinos' [10], Waxholm Space [4] and the Allen Reference Atlas [3], have some limitations. First, they do not have the desired level of details in the brainstem. Second, they are merely visual references. There are no tools for registering new data into the atlases. Third, they are not updateable. Fourth, they do not give the variance of structures. Lastly, a lot of manual work is involved in specifying registration landmarks and making annotations.

We propose a new type of atlas which we call the "active atlas". On the one hand, an active atlas is an anatomical reference model. It contains the mean and variance of position and shape of major anatomical structures. On the other hand, an active atlas includes landmark detectors. These detectors are texture classifiers trained in a semi-supervised fashion to detect regions with salient texture, such as nuclei and tracts, that can serve as registration landmarks. Such an atlas is "active" because the detectors allow our system to actively detect landmarks on new data for registration, instead of being a "passive" registration template; and also because these detectors can update themselves from new data, thus not "static".

This paper proposes a machine-learning based method for developing an active atlas, based on image series of Nissl-stained cryo-sectioned brains.

The rest of the paper is organized as follows. In Section 2 we describe related work. In Section 3 we describe the semi-supervised method we used to create landmark detectors. In section 4 we describe the alignment algorithm and the way in which we quantify the variation in the location of the landmarks. In section 5 we describe our experimental result. We conclude in section 6 with lessons learned and future plans.

2 Related work

[YC: this section needs revising]

Existing atlases of the mouse brain include Paxinos, Waxholm Space [4], and the more recent Allen Reference Atlas [3] and its successors, on top of which gene expression data [6, 8] as well as connectivity information [9] are mapped. The latest version of their common coordinate framework [1] is generated by deformably averaging two-photon microscopy images and manually demarcating structures with the help of immunohistochemistry data and genetic labeling data. Atlases for smaller organisms such as fruit flies [2, 11] and zebrafish [12, 13] are mostly based on volumetric images acquired by confocal microscopy. For registration, the BrainAligner program [11] relies on matching high-curvature points, and the ViBE-Z project [13] used trained classifiers to detect predefined 3D landmarks.

3 Methodology

3.1 Learning Texture Classifiers using Convolutional Neural Network

To bootstrap the atlas generation process, we picked an arbitrary stack, and asked an expert anatomist to annotate every other image for major landmarks by drawing polygons on the images using a homemade program (Figure 1). The results are 100 polygons for 19 different landmarks that span the entire brainstem, including nuclei (e.g. facial motor nucleus, trigeminal nucleus) and fiber tracts (e.g. trigeminal spinal tract, superior cerebellar peduncle). Patches of size 224-by-224 that are completely inside of each annotation polygon are extracted. Random patches from outside of any polygon are also extracted as patches for “background”. These patches form the training set for the texture classifier. A multiway classifier is obtained by fine-tuning the inception-BN network [1] using the training set. The network is pre-trained for ILSVRC2012 dataset.

For an input patch, the classifier outputs a 20-dimensional probability vector, indicating the probability that the patch belongs to either the background or each of the 19 landmarks.

The classifier is applied to images in all other stacks. To save computation, instead of predicting for patches centered at every pixel, the classifier predicts at grid points with a spacing of 56. The area between grid points is interpolated using bivariate spline. The result is a probability map for each image, where each pixel is assigned a probability vector.

3.2 Intra-stack Registration

As the brain sections are prepared using an advanced tape-transfer system, which almost perfectly preserves the shape of the sections, 3-parameter rigid transforms are sufficient to bring all sections into reasonable alignment. The transformations are found using the *elastix* program [5], based on maximizing the normalized correlation of the intensity between the thumbnail versions of the sections.

Once the registration transformations are known, a probability volume is reconstructed for each stack from the probability maps of all its sections. In a probability volume, each voxel has a probability vector. For the annotated stack, an annotation volume is reconstructed, in which each voxel has a scalar label. To facilitate registration at the next stage, the 3D contours of annotations are smoothed. We assume the sections all have constant thickness and are parallel to other sections in the same stack.

3.3 3D-3D Registration

To generate the atlas, we need to register the reconstructed volumes of all individuals in 3D and obtain an average. This process is done iteratively. We begin by using the annotated volume as the template and register the other volumes to it. Because the sections may be cut at different angles for different stacks, in effect resulting in shears between the volumes, we find 12-parameter 3D affine transformations.

Given the annotated volume Ω and a probability volume S , the best transformation is one that maximizes the “same-class overlap” objective:

$$M(T) = \sum_k \sum_{p \in \Omega_k} S_k[Tp] ,$$

where T is a 3-by-4 affine transform matrix, Ω_k is the set of voxels in the annotation volume that have label k , S_k is the volume formed by taking the entries corresponding to class k from the probability vectors of every voxel in S .

The optimization starts with grid search over the 3 translation parameters. Then gradient descent with Adagrad is used to approach the optimum.

3.4 Localizing Landmarks on Registered Images

To compute the statistics of landmarks’ position and shape, we need to accurately localize the landmarks of the registered volumes in 3D.

The fact that the probability volumes are registered to the annotation volume means the annotations can be propagated to these volumes. Figure 1 shows the contours of projected annotations on one of the registered images. These contours are not adapted to the actual images, but provide a rough guess for the landmarks’ positions. Starting from them, morphological snake [7] is used to fit the probability map of each landmark, giving more accurate contours.

After a small amount of human editing, these contours essentially become annotations for these registered stacks. Patches can be extracted from them to improve the texture classifier. This automatic augmentation of the training data constitutes the semi-supervised scheme of iteratively learning the landmark detectors.

3.5 Generating the Atlas

The atlas includes the statistics of the centroid of each landmark, as well as the variation modes of its shape.

Based on the transformations computed previously, landmark contours in registered images are reconstructed as 3D surface meshes in the common coordinate space defined by the annotation volume. The meshes are decimated and smoothed.

In each of the 9 brains, a landmark has two instances, one in each hemisphere. This gives 18 instances of each landmark for learning a 3D shape model of it.

4 Experiment Results

4.1 Accuracy of 3D Registration

4.2 Accuracy of Snake-adjusted Contours

4.3 Improvement of Classifier by Training Data Augmentation

4.4 Position and Shape Statistics of Landmarks

References

1. Technical white paper: Allen mouse common coordinate framework. Tech. rep. (2015) [2](#)
2. Chiang, A.S., Lin, C.Y., Chuang, C.C., Chang, H.M., Hsieh, C.H., Yeh, C.W., Shih, C.T., Wu, J.J., Wang, G.T., Chen, Y.C., et al.: Three-dimensional reconstruction of brain-wide wiring networks in drosophila at single-cell resolution. *Current Biology* 21(1), 1–11 (2011) [2](#)
3. Dong, H.: Allen Reference Atlas: a digital color brain atlas of the C57BL/6J male mouse. John Wiley & Sons (2008) [1](#), [2](#)
4. Johnson, G.A., Badea, A., Brandenburg, J., Cofer, G., Fubara, B., Liu, S., Nissanov, J.: Waxholm space: an image-based reference for coordinating mouse brain research. *Neuroimage* 53(2), 365–372 (2010) [1](#), [2](#)
5. Klein, S., Staring, M., Murphy, K., Viergever, M.A., Pluim, J.P.: Elastix: a toolbox for intensity-based medical image registration. *Medical Imaging, IEEE Transactions on* 29(1), 196–205 (2010) [3.2](#)
6. Lein, E.S., Hawrylycz, M.J., Ao, N., Ayres, M., Bensinger, A., Bernard, A., Boe, A.F., Boguski, M.S., Brockway, K.S., Byrnes, E.J., et al.: Genome-wide atlas of gene expression in the adult mouse brain. *Nature* 445(7124), 168–176 (2007) [2](#)
7. Marquez-Neila, P., Baumela, L., Alvarez, L.: A morphological approach to curvature-based evolution of curves and surfaces. *Pattern Analysis and Machine Intelligence, IEEE Transactions on* 36(1), 2–17 (2014) [3.4](#)
8. Ng, L., Bernard, A., Lau, C., Overly, C.C., Dong, H.W., Kuan, C., Pathak, S., Sunkin, S.M., Dang, C., Bohland, J.W., et al.: An anatomic gene expression atlas of the adult mouse brain. *Nature neuroscience* 12(3), 356–362 (2009) [2](#)
9. Oh, S.W., Harris, J.A., Ng, L., Winslow, B., Cain, N., Mihalas, S., Wang, Q., Lau, C., Kuan, L., Henry, A.M., et al.: A mesoscale connectome of the mouse brain. *Nature* 508(7495), 207–214 (2014) [2](#)
10. Paxinos, G., Franklin, K.B.: The mouse brain in stereotaxic coordinates. Gulf Professional Publishing (2004) [1](#)

11. Peng, H., Chung, P., Long, F., Qu, L., Jenett, A., Seeds, A.M., Myers, E.W., Simpson, J.H.: Brainaligner: 3d registration atlases of drosophila brains. *Nature methods* 8(6), 493–498 (2011) [2](#)
12. Randlett, O., Wee, C.L., Naumann, E.A., Nnaemeka, O., Schoppik, D., Fitzgerald, J.E., Portugues, R., Lacoste, A.M., Riegler, C., Engert, F., et al.: Whole-brain activity mapping onto a zebrafish brain atlas. *Nature methods* 12(11), 1039–1046 (2015) [2](#)
13. Ronneberger, O., Liu, K., Rath, M., Rueß, D., Mueller, T., Skibbe, H., Drayer, B., Schmidt, T., Filippi, A., Nitschke, R., et al.: Vibe-z: a framework for 3d virtual colocalization analysis in zebrafish larval brains. *Nature Methods* 9(7), 735–742 (2012) [2](#)